

## GROWTH AND SURVIVAL OF POSTLARVAL PENAEUS AZTECUS UNDER CONTROLLED CONDITIONS OF TEMPERATURE AND SALINITY<sup>1</sup>

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Temperature and salinity may be considered among the most important abiotic factors influencing the growth and survival of much of the estuarine fauna. They are of particular significance to those organisms that spend certain portions of their life cycle in the open sea where both factors are relatively stable, and other portions in the estuarine areas where both temperature and salinity may change drastically. Although temperature is generally thought to overshadow salinity in its effects on migratory organisms, salinity, probably through its osmotic effects, also plays a part in limiting some organisms to specific environments.

Several investigators have attempted to evaluate the importance of temperature and salinity to penaeid shrimp of the Gulf of Mexico, but ecological questions concerning these factors remain unanswered. Although field studies have dealt with the relationship of shrimp to salinity, the conclusions reached have differed widely enough to warrant further investigation. The interpretation of observations on salinity and shrimp abundance in nature is made difficult by changes in other environmental factors, some of which frequently vary with salinity. Such factors include temperature, light, substrate, food supply, cover and pollution. For this reason, controlled-environment studies in the laboratory were employed in the present work.

In an earlier study, Zein-Eldin (1963) determined that under conditions of constant temperature and somewhat restricted food supply, grooved *Penaeus* post-larvae<sup>2</sup> survived and grew over a wide range of salinity (2-40‰). However, it has been suggested that in other migratory Crustacea, notably in the European shrimp, *Crangon crangon* (Broekema, 1941), as well as in juvenile and adult brown and pink shrimp, *Penaeus aztecus* and *P. duorarum*, respectively (Williams, 1960), temperature can influence tolerance to salinity. Thus, further studies were designed to test the combined effects of temperature and salinity on the survival and growth of postlarval brown shrimp.

### MATERIALS AND METHODS

The work was of two types, 24-hour survival studies and a 28-day growth experiment. For all work, postlarval brown shrimp of approximately 12 mm. rostrum-telson length were seined from the Gulf of Mexico surf at the entrance to Galveston Bay. The animals were held in the laboratory in aerated water of ap-

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<sup>2</sup> As defined by Renfro (1964).

proximately 25‰ and 25° C. for at least 24 hours prior to use. Few mortalities occurred during this preliminary holding period.

The first objective was to obtain a rapid, crude estimate of postlarval tolerance to salinity and temperature in order to provide guidelines for the more detailed and sensitive growth study to follow. Accordingly, we selected short-term survival as a rough index suited to our needs.

To determine the short-term tolerance of brown shrimp to salinity and temperature, we exposed groups of experimental shrimp to different levels of the two factors for 24 hours. The test levels were chosen to include and extend above and below the ranges of salinity and temperature at which large numbers of postlarvae have been observed in nature (Bearden, 1961; Williams, 1955; Baxter, unpublished).

Temperature control of  $\pm 0.5^\circ$  C. was maintained by B.O.D.-type incubators. Salinity changes were effected by replacing portions of water in the test containers with equal volumes of either distilled water or evaporation-concentrated sea water. Salinity was determined by hydrometer and reported to the nearest part per thousand. Four series of 24-hour survival experiments were carried out with groups of 5 to 30 animals as described below. Series 1, 2 and 4 had an initial salinity of 24–25‰ and an initial temperature of 24° to 26° C., matching conditions of the holding aquaria. Initial salinity in Series 3 was 40‰, equal to the unusually high level at which animals for that series were collected. Following introduction of the shrimp into the vigorously aerated experimental containers, stepwise changes in both temperature and salinity (0 to 8 steps, depending on the magnitude of change involved) were made over a 10- to 12-hour period, to reach the desired conditions. The attainment of these conditions marked the beginning of the 24-hour test period. At the end of that time, the beakers were removed from the incubators and the live and dead postlarvae counted. Failure to show either spontaneous or probe-induced activity upon return to room temperature was considered indicative of death. No food was provided during the experiment.

Previous observations of postlarvae in the laboratory had indicated that failure to keep them under water mechanically could lead to considerable mortality, due to their jumping activity. This type of loss was avoided during the first two series by restraining each group of animals in a one-liter beaker whose mouth was filled with a stemless funnel 4 inches in diameter. When the beaker was filled with water, all air space accessible to the shrimp was eliminated, thereby preventing the animals from escaping the vessel or adhering to its dry surfaces. Aeration was provided by means of an air stone attached to  $\frac{3}{16}$ -inch O.D. Tygon tubing fitted through the hole in the funnel.

In the first two series, we employed only five shrimp per group, hoping thus to reduce cannibalism. Although losses caused by escape from the water were successfully avoided, test results indicated that cannibalism occurred at intermediate and higher temperatures where the number of survivors plus the number of dead animals remaining per group frequently totaled less than the original number of shrimp (Table I). In such cases, we attempted to discriminate between deaths due to cannibalism and mortalities attributable to salinity and temperature by arbitrarily assuming that salinity-temperature combinations causing the rapid death of some experimental shrimp were sufficiently severe to inhibit feeding, including cannibal-

TABLE I

*Survival of P. aztecus postlarvae after 24-hour exposure to various combinations of salinity and temperature*

Series no.	Shrimp per group	Acclimated for: (hrs)	Experimental conditions																										
			7					15					25					30					35						
1	5	10-12	Temperature (° C.)	3	8	23	32	36	5	10	24	34	37	5	11	25	34	37	6	10	26	36	41	7	11	27	36	37	
			Salinity (‰)																										
			Survivors	0	1	5	5	4	1	4	5	5	5	3	4	5	5	5	4	4	4	5	4	1	2	1	1	3	
2	5	10-12	Intact dead*	5	4	0	0	1	4	1	0	0	0	0	0	0	0	0	0	1	0	0	0	4	1	2	3	1	
			Temperature (° C.)																										
			Salinity (‰)	5	9	24	34	38	4	9	24	34	38	4	8	24	34	38	4	9	24	33	37	4	9	25	34	39	
3	10	10-12	Survivors	0	2	5	5	4	0	3	5	5	5	4	4	5	5	4	3	3	5	4	5	0	1	1	3	2	
			Intact dead*	5	3	0	0	1	5	2	0	0	0	0	0	0	1	0	0	0	0	0	0	5	3	4	0	2	
			Temperature (° C.)																										
4	30	10-12	Salinity (‰)	6	10	25	36	37	7	11	26	34	38	7	10	26	35	39	6	11	26	34	39	7	11	26	35	39	
			Survivors	0	6	10	10	10	8	10	10	10	10	8	10	10	10	10	10	10	10	10	10	10	9	9	10	10	10
			Temperature (° C.)																										
—	30	0	Salinity (‰)																										
			Survivors																										
			Temperature (° C.)																										
—	30	0	Salinity (‰)																										
			Survivors																										
			Temperature (° C.)																										

\* These were dead shrimp which showed no signs of having been eaten by the other experimental animals within the group. See text.



ism, among the survivors. High rates of penaeid activity, including movement, feeding, and molting, have been observed at temperatures of 25° and 32° C. in this laboratory. Such activities favor cannibalism among shrimp under relatively crowded experimental conditions. On this basis, we counted as mortalities only those dead animals visibly present at the end of each 24-hour experimental period.

To test this assumption, we required data unaffected by cannibalism. These were obtained in Series 3 by confining each animal within a 1½-inch length of 14-mm. Pyrex tubing, both ends of which were covered with cotton gauze held in place with small rubber bands. While permitting contact between the animal and its experimental aquatic environment, this procedure prevented "jump-out" losses and made physical contact between shrimp impossible. Ten postlarvae thus isolated were placed in each test beaker and the experimental conditions established as before. The survival results agreed well with those of the two earlier series as interpreted above, tending to substantiate our assumption regarding the effect of cannibalism.

TABLE II

*Schedule of salinity and temperature changes [initial salinity and temperature were 26‰ and 23° C., respectively]*

Elapsed time (hr.)	Desired salinity (‰)					Desired temperature (° C.)			
	2	5	15	25	35	11	18	25	30
	Actual salinity					Actual temperature			
2	20	20	20	25	25	23	23	23	26
8	15	15	15	25	30	19	22	24	30
24	10	10	15	25	35	17	19	25	33
36	5	5	15	25	35	12	18	25	33
48	2	5	15	25	35	11	18	25	32

In a fourth series, further survival data were sought at temperature-salinity combinations which seemed to be near the extremes of postlarval tolerance, as suggested by results of the three previous series (Table I). In this series the importance of acclimation was also estimated. Each set of experimental conditions was duplicated in two two-liter beakers, one for shrimp acclimated as usual, the other for animals which were transferred directly from the holding tank to the extreme salinity and temperature levels to be tested. Thirty individually confined postlarvae were held in each beaker, and 24-hour survival determined as before.

For the growth study, 46 liters of brackish water and 100 animals were placed in each of twenty 15-gal. aquaria. Filtration, aeration, and confinement were accomplished as previously described (Zein-Eldin, 1963). Five aquaria were placed in each of four constant-temperature rooms. The experimental temperatures were changed from the initial 23° C. to 11°, 18°, 25°, or 32° C. Water temperature, although  $\pm 0.5^\circ$  C. in a given aquarium, varied as much as 1° C. among aquaria in a single room. The initial salinity of 23‰ was simultaneously adjusted stepwise with temperature, to final levels of 2‰, 5‰, 15‰, 25‰, or 35‰ (Table II). Each tank was continuously illuminated by two 40-w. fluorescent lamps.

Postlarvae were fed live brine shrimp (*Artemia*) nauplii throughout the growth experiment. The nauplii in a 0.1-ml. sample of brine shrimp in water were counted to estimate number per unit volume, and the volume of food recorded at each feeding of the postlarvae. *Artemia* nauplii were filtered and washed with distilled water before their addition to the tanks, in order to avoid increases in experimental salinity levels. Live brine shrimp nauplii were present in excess in all aquaria during the first 24 days of the experiment. During the last four days at 32° C., however, the shrimp had grown to such a size that maintaining an excess food supply became almost impossible, even though 400,000 to 500,000 nauplii per tank (a minimum of 9000 to 10,000 per experimental animal) were supplied per day.

TABLE III  
Cumulative mortality [only observed deaths are included]

Elapsed time (days)	Temperature (° C.)	Salinity (‰)	32					25					18					11				
			2	5	15	25	35	2	5	15	25	35	2	5	15	25	35	2	5	15	25	35
1			5	1	0	0	0	0	0	0	0	0	1	2	0	0	0	0	0	0	0	0
2			41	25	16	2	0	0	0	0	0	0	2	2	0	0	0	4	2	0	0	0
3			44	25	16	6	0	0	0	0	0	0	13	3	0	0	0	77	2	0	0	0
4			45	25	17	7	0	0	0	0	0	0	17	3	0	0	0	93	5	0	0	0
5			45	25	17	7	0	0	0	0	0	0	19	3	0	0	0	97	12	0	0	0
6			45	25	17	7	0	0	0	0	0	0	19	3	0	0	0	97	12	0	0	0
7			45	25	17	7	0	0	0	0	0	0	19	3	0	0	0	97	22	0	0	0
8			45	25	17	7	0	0	0	0	0	0	19	3	0	0	0	97	25	0	0	0
9			45	25	17	7	0	0	0	0	0	0	19	3	0	0	0	97	32	0	0	0
10			45	25	17	7	0	0	0	0	0	0	19	3	0	0	0	97	37	0	0	0
11			45	25	17	7	0	0	0	0	0	0	24	3	0	0	0	97	41	0	0	0
12			45	25	17	7	0	0	0	0	0	0	26	4	0	0	0	97	45	0	0	0
13			45	25	17	7	0	0	0	0	0	0	27	4	0	0	0	97	50	0	0	0
14			45	25	17	7	0	0	0	0	0	0	27	4	0	0	0	97	55	0	0	0
17			45	25	17	7	0	0	0	0	0	0	29	4	0	0	0	97	60	0	0	0
18			45	25	18	7	0	0	0	0	0	0	31	4	0	0	0	97	60	0	0	0
19			45	25	18	7	0	0	0	0	0	0	45	4	0	0	0	97	60	0	0	0
20			46	25	18	7	0	23	0	0	0	0	57	4	0	0	0	97	63	0	0	0
21			46	25	18	7	0	39	0	0	0	0	58	5	0	0	0	97	63	0	0	0
22			46	25	18	7	0	50	0	0	0	0	58	5	0	0	0	97	63	0	0	0
24			46	25	18	7	0	55	0	0	0	0	58	5	0	0	0	97	66	0	0	0
28			46	25	18	7	2	55	0	0	0	0	58	5	0	0	0	97	67	0	0	4
No. of animals removed for measurement			25	30	35	50	50	40	50	50	50	50	36	50	50	50	50	0	30	50	50	50
No. escaped			2	0	0	0	0	4	4	3	0	0	1	2	2	2	0	0	0	0	0	0
Observed survivors			15	22	25	29	47	0	43	43	50	49	0	41	46	48	49	0	0	49	46	43
Unobserved deaths			12	23	22	14	1	1	3	4	0	1	5	2	2	0	1	3	3	1	4	3
Per cent survival			21	31	38	58	94	0	93	91	100	98	0	85	96	100	98	0	0	98	92	86

At approximately 5-day intervals, 10 animals were removed from each aquarium. These included both the largest and smallest specimens, and eight collected at random. The animals were individually measured to the nearest 0.5 mm., blotted dry, weighed to the nearest 0.1 mg. with a Mettler H15 analytical balance, and preserved. At the termination of the experiment, all remaining animals were similarly treated. The final per cent survival was determined by comparing the total number of shrimp remaining at the close of the experiment to the number that theoretically should have been present (original number less those that had been removed for sampling and a few that had escaped). The unobserved deaths recorded in Table III were animals not accounted for either as survivors, observed deaths, or those sampled for measurement.

On the assumption that an individual *Artemia* nauplius weighs an average of 7.1  $\mu$ g. (D. Godwin, unpublished), we estimated conversion efficiency by comparing the calculated wet weight of the brine shrimp that were fed with the weight



gain of the surviving penaeids. Determinations were made only for those temperature and salinity combinations at which survival was 85% or greater.

Although the design of the experiment was similar to that of Costlow, Bookhout and Monroe (1960, 1962) in studies of larval crab survival, we did not use the statistical methods which they employed. The fitted-surface method of Box and Youle (1955) has proved valuable in industrial applications of physical and chemical interactions whose principles are sufficiently well defined to permit relatively safe

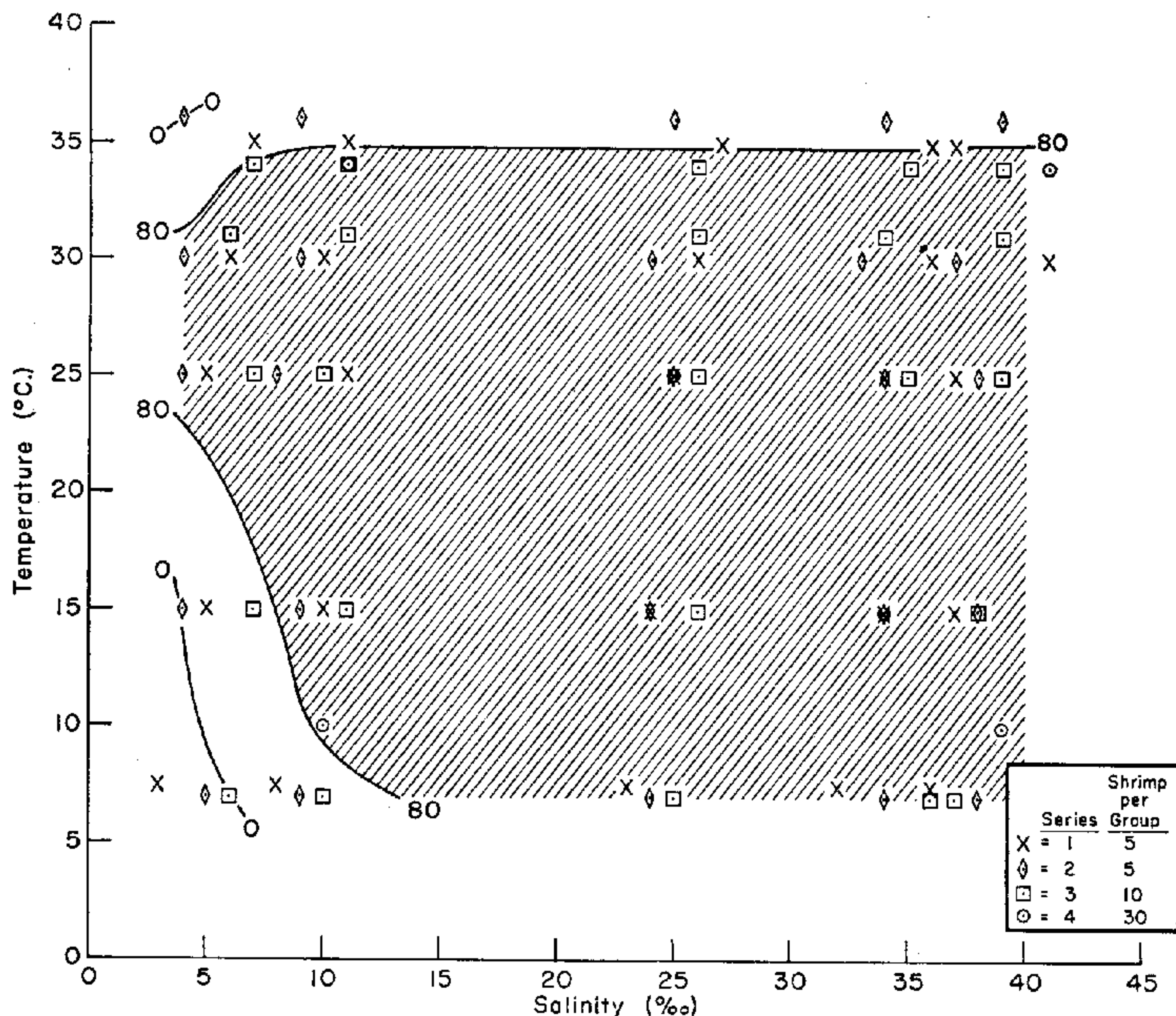


FIGURE 1. Per cent survival of *P. aztecus* postlarvae after 24 hours at indicated levels of salinity and temperature.

extrapolation from a limited number of experimental observations. However, the complex nature of biological responses to temperature and salinity renders such extrapolation extremely speculative. In the present study, we have tested a group of temperature-salinity combinations which represents a relatively large range of levels for each factor. Our interpretations of the results exclude extrapolation.

## RESULTS AND DISCUSSION

### Short-term survival

The excellent survival of postlarvae for periods of 24 hours under most of the experimental conditions suggested a broad zone of short-term tolerance to both

salinity and temperature (Table I). The animals were quite euryhaline, especially at 25° and about 30° C., although a marked reduction in tolerance to salinity levels below 10‰ was demonstrated at 7° and 15° C. (Fig. 1). A general reduction in survival near 35° C., regardless of salinity, suggests a strong temperature effect. The absolute limiting (maximum) temperature for *P. aztecus* is probably only slightly above 35° C.

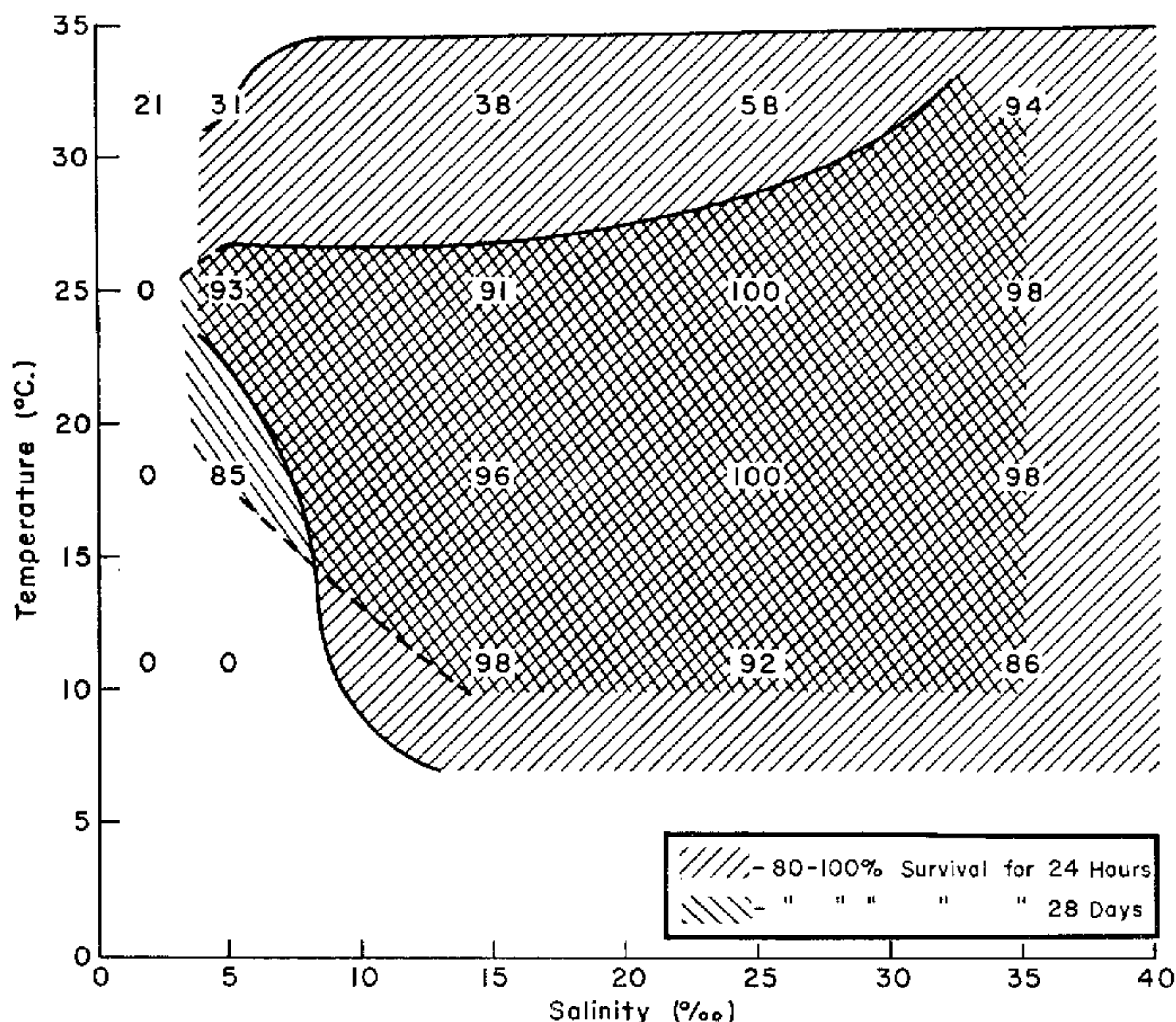
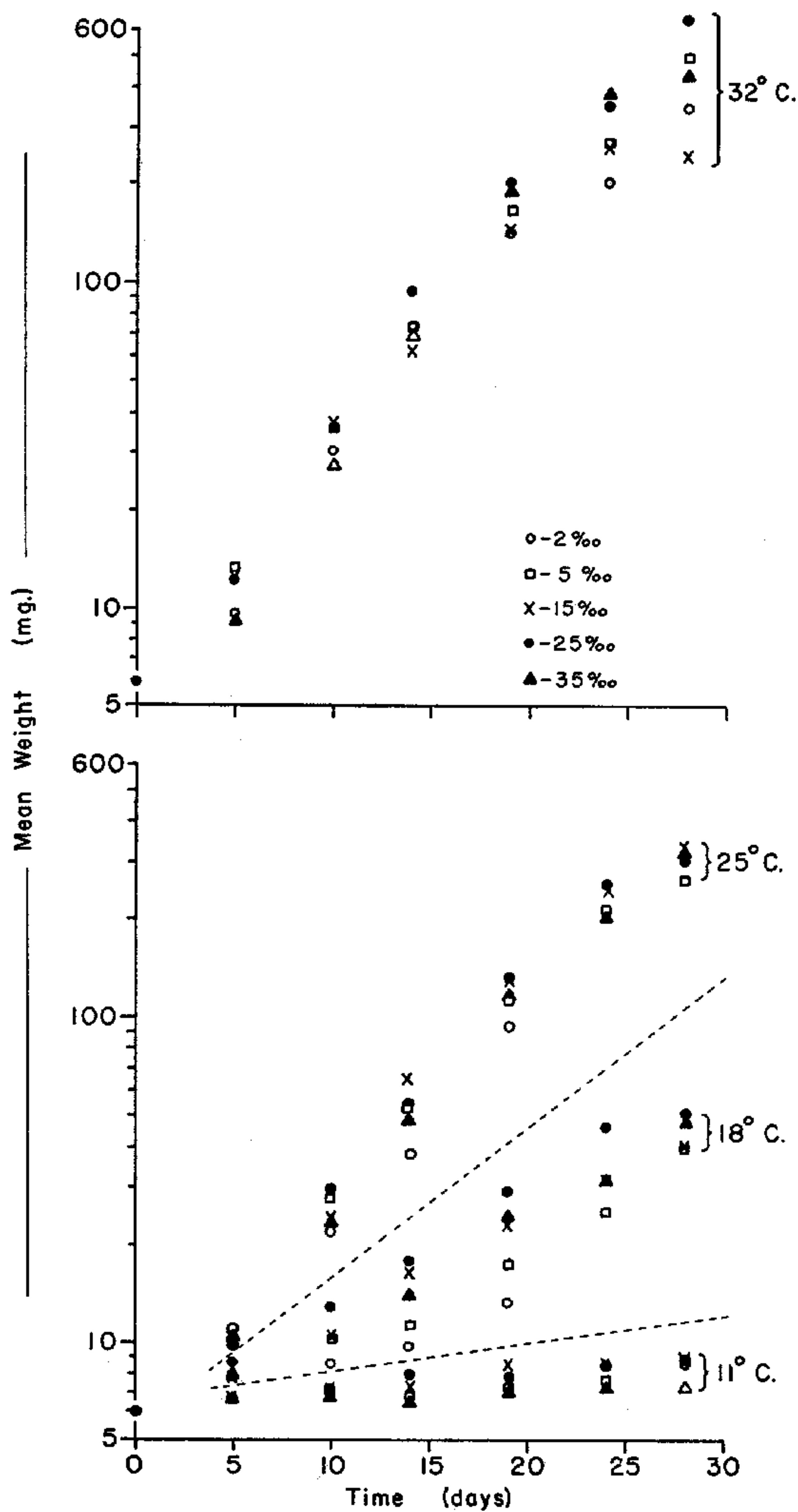


FIGURE 2. Long- and short-term survival of *P. aztecus* at indicated levels of salinity and temperature. Numbers indicate 28-day survival in per cent.

The effect of acclimation in extending ranges of postlarval tolerance is clearly shown in the results of the fourth series. In each of the four combinations of temperature and salinity, gradually changed conditions permitted better survival than did sudden changes (Table I). This effect was considerably more marked at 10° than at 35° C.

#### 28-Day survival

The survival of postlarvae in the 28-day experiment further confirmed this wide zone of tolerance to salinity and temperature. Although the per cent survival for 28 days was somewhat lower than that observed for only 24 hours, in most cases the results were much the same (Fig. 2). There is some suggestion of greater long-term survival for animals at low temperature and low salinity than

FIGURE 3. Growth of young *P. aztecus* at indicated levels of temperature and salinity.



for those exposed only 24 hours. This apparently paradoxical situation is probably related to the longer acclimation period employed in the 28-day study.

Survival was markedly reduced at the highest temperature (32°C.) at all salinities tested except 35‰. Much of the accountable mortality at this temperature occurred during the first four days (Table III) and was presumably the result of the immediate stress caused by the changes in environment. However, the stress of salinity acclimation would not seem to explain the poorer survival observed at 25‰ (very near the initial salinity) than at 35‰. The relatively large numbers of unobserved deaths occurring at 32° C. and 2‰, 5‰, 15‰ and 25‰ (Table III) suggest two other possible causes of mortality—the experimental temperature *per se*, and increased cannibalism associated with high temperatures (as noted above in Series 1 and 2 of the 24-hour studies). It is possible that at 32° C. the one-month period of exposure in the growth experiment elicited long-term temperature effects which could not be manifested in the relatively short duration of the 24-hour studies.

TABLE IV

*Increase in mean length (mm.) of P. aztecus surviving 28 days  
at indicated levels of temperature and salinity*

Temperature (° C.)	Salinity (‰)				
	2	5	15	25	35
32	23.4	28.9	19.0	32.0	25.8
25	—	21.9	24.4	22.3	22.6
18	—	6.3	6.5	7.4	7.6
11	—	—	0.5	0.5	0.4

Mortalities occurring at other temperatures were limited to the lowest salinities, with stress due to reduced salinity and temperature being sufficient to kill all the animals at 2‰ and 11° C. in only 5 days (Table III). The mortalities observed at 5‰ and 11° C., as well as those at 2‰ and 18° C., probably reflect the cumulative effects of stress, since deaths occurred continuously throughout the course of the experiment. At 25° C. and 2‰, however, all observed deaths occurred during a four-day period late in the experiment. Although the initial cause of this mortality is not known, later deaths (on the 21st through the 24th day) were probably due to fouling of water, since brine shrimp were also dying. Furthermore, an earlier experiment (Zein-Eldin, 1963) had indicated that *P. aztecus* postlarvae survive well under these conditions.

### Growth

Differences in rate of growth were more closely related to temperature than to salinity (Fig. 3). The relative effects of the two factors may be readily determined by comparing the magnitude of growth differences associated with variation in salinity (columns) with that due to variation in temperature (rows) (Table IV). Differences in mean length between temperature groups were detectable as early as the first sampling period (5 days) and increased in magnitude during the experi-

mental period (Fig. 4). Both length and weight increased much more rapidly at 32° and 25° C. than at lower temperatures. The maximum increase in size was observed at 32° and 25‰, conditions under which one animal grew to 50 mm. and 962 mg., a more than four-fold increase in length and a weight increase of 150-fold (Table V). The great variation in size noted earlier (Zein-Eldin, 1963) was also observed in this experiment, with differences in length between smallest and largest

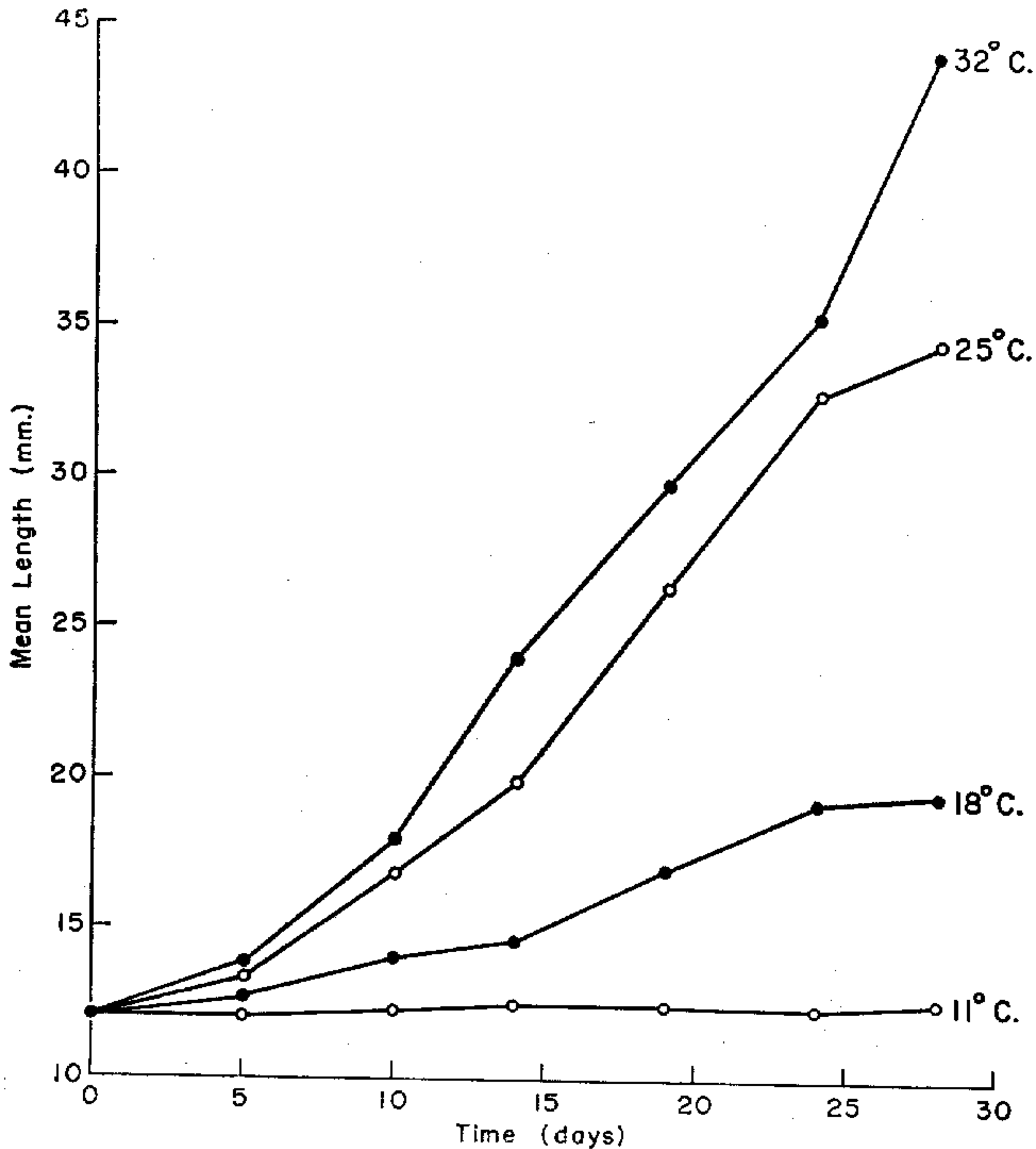


FIGURE 4. Growth of young *P. aztecus* at various temperatures (salinity: 25‰).

animals ranging from 13 to 25.5 mm. in the various salinities at 25° and 32° C. (Table V). Almost no growth was detected at 11° C., although survival was good at salinities of 15‰ and above. With the exception of 32° C. and corresponding salinity levels of 25‰ and below, where mortality more than offset the rapid growth rate (Tables IV and VI), the gain in total weight of the survivors was comparable within a temperature. However, this gain increased approximately 10-fold within each level of salinity between 11° and 18° C., and only slightly less between 18° and 25° C. (Table VI).

TABLE V

Mean size and growth rate of growth-experiment survivors, including 10 animals sampled at 28 days and shown in Table VII. Size range given in parentheses. Initial weight and length were 6.1 mg. and 12.1 mm., respectively

Temperature and salinity	Number of survivors	Weight (mg.)	Length (mm.)	Increase in length per day (mm./day)
32° C.				
2‰	15	340.6 (157.7–735.1)	34.8 (27.5–47.5)	0.81 (0.55–1.26)
5‰	22	447.6 (241.0–667.6)	39.6 (33.0–46.0)	0.98 (0.75–1.21)
15‰	25	240.2 (35.2–542.0)	31.0 (17.0–42.5)	0.68 (0.18–1.09)
25‰	29	610.8 (309.0–961.9)	43.1 (36.0–50.0)	1.11 (0.85–1.35)
35‰	47	423.7 (164.7–753.6)	38.3 (28.5–46.5)	0.94 (0.59–1.23)
25° C.				
5‰	43	274.0 (115.3–482.0)	33.9 (25.0–41.5)	0.77 (0.46–1.05)
15‰	43	375.3 (163.8–681.1)	37.4 (29.0–46.5)	0.90 (0.60–1.23)
25‰	50	313.8 (101.4–538.0)	35.2 (24.0–44.0)	0.82 (0.42–1.14)
35‰	49	291.0 (108.0–605.7)	34.4 (24.5–43.0)	0.80 (0.44–1.10)
18° C.				
5‰	41	33.9 (18.0–56.2)	18.3 (14.0–21.5)	0.20 (0.08–0.34)
15‰	46	43.7 (15.3–77.7)	19.1 (14.5–23.0)	0.25 (0.09–0.39)
25‰	48	52.8 (15.0–101.4)	20.1 (14.0–25.0)	0.29 (0.08–0.46)
35‰	49	35.2 (17.7–62.7)	18.3 (14.5–22.0)	0.22 (0.09–0.35)
11° C.				
15‰	49	8.7 (5.2–13.1)	12.6 (11.0–14.0)	0.03 (0–0.08)
25‰	46	8.8 (6.4–10.8)	12.7 (11.5–14.0)	0.02 (0–0.08)
35‰	43	7.6 (5.5–9.7)	12.4 (11.0–13.0)	0.01 (0–0.08)

Growth rate based only upon the steepest portions of the growth curves (*i.e.*, between the 10th and 28th days) approached a value of 1.4 mm. per day at 32° C. and 1.1 mm. per day at 25° C., as against the lower values of 1.1 mm. per day at 32° and 0.8 mm. per day at 25° C. over the entire experimental period (Fig. 4, Table VII). Although the mean growth rates reported here (Table V) for both 25° and 32° C. exceed the maximum of 0.56 mm. per day which Pearson (1939) reported for laboratory-held postlarvae of *P. brasiliensis* (probably *P. aztecus*), and the maximum of 1.35 mm. per day far exceeds his value, these rates do not

TABLE VI

Increase in total weight (g.) of *P. aztecus* surviving 28 days at indicated levels of temperature and salinity. Food conversion efficiency (%) indicated in parentheses where survival was 85% or greater

Temperature (° C.)	Salinity (‰)				
	2	5	15	25	35
32	4.7	9.6	5.8	17.5	19.6 (37)
25	—	11.5 (43)	15.9 (53)	15.4 (46)	14.0 (43)
18	—	1.2 (32)	1.7 (33)	2.2 (40)	1.6 (34)
11	—	—	0.1 (12)	0.1 (14)	0.1 (5)



approach that of postlarval white shrimp, *P. setiferus*, which grew an average of 2.1 mm. per day in pond experiments conducted by Johnson and Fielding (1956). A similarly low rate of growth for aquarium-held *Metapenaeus mastersii*, which ranged in carapace length from 1.9 to 7.0 mm., has been reported by Dall (1958). Laboratory animals grew only 10 mm. in total length per month at 24° to 28° C., as against a natural growth rate of 20 to 30 mm. per month.

Growth of our laboratory-held *P. aztecus* postlarvae likewise only approached that of slightly larger animals in the field. Viosca (1920) estimated a growth rate of approximately 25 mm. per month for *P. setiferus* in the length range 30 to 150 mm., while Gunter (1950) observed a rate of 25 to 40 mm. per month for the same species growing from 28 to 100 mm. Extrapolation of Lindner and Anderson's (1956) growth curve for white shrimp gives a rate of 1.50 mm. per day for shrimp between 20 and 65 mm. in length. Williams (1955) determined a rate of 1.7 mm. per day for *P. aztecus* growing from 37 to 102 mm., a rate also estimated by St. Amant, Corkum and Broom (1963) for 51- to 125-mm. specimens of this species. It must be noted, however, that these estimates were for shrimp at the upper end of the size ranges encountered in the studies described here.

Although the studies of conversion efficiency were necessarily crude, the resulting data indicated that the most efficient utilization of food occurred at 25° C. (Table VI). Because of the high mortality, no efficiencies could be calculated for shrimp held at 32° C. in salinities of 25‰ and lower. The conversion values should be considered maximal at the highest temperatures since some cannibalism probably occurred. However, efficiencies for the animals held at 11° C. are probably low since it was apparent that much of the food provided was not eaten. At this low temperature, the postlarvae were generally inactive, resting most of the time on the bottom.

Johnson and Fielding (1956), studying *P. setiferus* in aquaria during August (temperature not stated), found a mean food-conversion efficiency of 19% for juveniles (mean weight 0.9 to 2.1 g.) held one week at 18.5‰, as against an efficiency of 24% for juveniles (mean weight 0.7 to 2.1 g.) held one week at 34‰. All animals were fed at a rate of 10% of the initial body weight per day, a rate less than that provided in our experiments. The lower efficiencies determined by Johnson and Fielding may be due, in part, to the larger size of the animals held, since it has been shown in fishes (Kinne, 1960) that conversion efficiency decreases with increasing size. Furthermore, Johnson and Fielding obtained the maximum efficiency of 50% from a group of animals of 0.7 g. mean weight held at 34‰. This value compares favorably with those reported here for brown shrimp of mean weight 0.3 g. and less, and would indicate that rapidly growing young shrimp require 2 to 4 g. of utilizable food to produce 1 g. of tissue.

The decrease in growth rate of animals held at 32° C. and 15‰ is unexplainable. This group of animals consumed less food during the latter days of the experiment, even though excess food was present. If this decreased growth represented a long-term effect of the combined stresses of lowered salinity and increased temperature, it is strange that such a decrease did not occur among groups held at even lower salinities at this temperature. Interpretation of the combined effects of lower salinity with 32° C. on growth was complicated by the high rates of mortality among these groups.

TABLE VII

Mean animal weight (mg.) and length (mm.) at each salinity and temperature level. All individuals initially 6.1 ( $\pm 0.2$ ) mg. and 12.1 ( $\pm 0.2$ ) mm. Values are based on samples of 10 except those that are starred, which are based on samples of 5. The largest and smallest remaining animals in each test group were included in each sample. Figures in parentheses indicate one standard error

Temp. (° C.) and elapsed time (days)	Level of salinity (‰)									
	2		5		15		25		35	
	Weight	Length	Weight	Length	Weight	Length	Weight	Length	Weight	Length
32°										
5	9.7* (0.9)	13.1* (0.3)	13.5 (1.0)	14.0 (0.4)	12.4 (0.9)	13.8 (0.3)	12.1 (1.0)	13.8 (0.3)	9.3 (0.6)	12.8 (0.3)
10	31.1* (4.9)	17.4* (0.3)	35.9 (3.8)	17.6 (0.6)	37.8 (7.8)	17.8 (1.0)	36.8 (5.8)	18.0 (0.9)	28.1 (4.3)	16.6 (0.7)
14	74.7* (15.8)	22.3* (1.5)	75.8* (12.0)	23.0* (1.2)	62.2 (10.9)	20.8 (1.0)	94.6 (13.0)	24.0 (1.2)	70.2 (9.8)	21.5 (1.0)
19	144.5* (11.2)	27.1* (2.3)	167.3* (16.6)	28.3* (0.9)	148.2* (33.6)	26.6* (2.2)	206.5 (34.7)	29.8 (2.0)	195.8 (23.0)	30.1 (1.2)
24	209.3* (61.0)	30.0* (2.9)	271.3 (32.0)	33.4 (1.2)	265.1* (59.5)	32.2* (2.6)	356.8 (53.7)	35.4 (1.9)	379.6 (51.5)	36.4 (1.6)
28	347.7 (54.1)	35.6 (1.9)	482.3 (23.0)	41.0 (0.6)	246.9 (53.6)	31.1 (2.5)	654.0 (73.8)	44.1 (1.7)	437.2 (57.8)	38.0 (1.8)
25°										
5	9.7 (0.7)	13.1 (0.3)	10.4 (0.7)	13.4 (0.2)	10.8 (0.5)	13.3 (0.2)	11.1 (0.6)	13.4 (0.3)	10.5 (0.6)	13.0 (0.2)
10	21.8 (2.0)	15.9 (0.5)	27.9 (3.0)	17.0 (0.5)	23.0 (3.1)	15.8 (0.6)	29.9 (4.6)	16.8 (0.8)	23.0 (2.2)	16.1 (0.5)
14	37.9 (3.4)	18.4 (0.6)	51.1 (8.6)	20.0 (1.1)	64.7 (12.5)	21.2 (1.3)	54.0 (8.4)	19.9 (1.0)	48.3 (5.1)	19.6 (0.7)
19	93.2 (9.6)	24.0 (1.3)	113.7 (20.6)	24.9 (1.4)	126.7 (22.4)	26.1 (1.3)	132.4 (17.1)	26.4 (1.1)	118.6 (16.9)	25.4 (1.4)
24	—	—	217.0 (25.6)	31.1 (1.2)	245.1 (24.9)	32.4 (1.2)	257.8 (27.0)	32.8 (1.1)	202.7 (27.8)	30.6 (1.2)
28	—	—	264.4 (27.4)	33.0 (1.2)	357.1 (41.8)	36.6 (1.4)	302.0 (41.2)	34.4 (1.7)	321.3 (44.3)	34.8 (1.3)
18°										
5	6.5 (0.2)	12.7 (0.1)	7.5 (0.2)	12.8 (0.1)	8.0 (0.4)	12.8 (0.2)	8.6 (0.5)	12.7 (0.2)	7.9 (0.2)	12.6 (0.1)
10	8.7 (0.4)	12.8 (0.2)	10.0 (0.1)	13.1 (0.1)	10.5 (0.6)	13.2 (0.2)	12.9 (0.8)	14.0 (0.2)	10.4 (0.6)	13.6 (0.3)
14	9.7 (0.5)	13.2 (0.2)	11.4 (0.8)	14.0 (0.3)	16.5 (1.6)	15.0 (0.5)	17.7 (1.6)	14.6 (0.5)	14.0 (1.0)	14.0 (0.3)
19	13.2* (1.1)	14.1* (0.2)	17.4 (1.0)	14.7 (0.2)	23.0 (1.7)	16.0 (0.1)	29.0 (3.6)	17.0 (0.7)	24.2 (2.1)	15.8 (0.3)
24	—	—	25.1 (2.7)	16.1 (0.6)	31.8 (2.9)	17.8 (0.5)	46.0 (4.7)	19.2 (0.6)	31.2 (3.6)	17.6 (0.5)
28	—	—	40.1 (4.0)	18.4 (0.7)	40.6 (4.9)	18.6 (0.7)	49.9 (7.7)	19.5 (1.0)	47.4 (4.0)	19.8 (0.7)
11°										
5	—	—	6.7 (0.3)	12.0 (0.2)	6.7 (0.3)	12.1 (0.1)	6.4 (0.3)	12.1 (0.1)	6.8 (0.2)	12.4 (0.1)
10	—	—	7.1* (0.3)	12.4 (0.2)	7.0 (0.3)	12.0 (0.2)	7.2 (0.3)	12.2 (0.2)	6.9 (0.4)	12.3 (0.2)
14	—	—	6.9* (0.3)	12.3 (0.1)	7.3 (0.2)	12.5 (0.2)	8.0 (0.3)	12.5 (0.2)	6.5 (0.3)	11.9 (0.2)
19	—	—	7.3* (0.4)	12.4 (0.1)	8.6 (0.3)	13.0 (0.2)	7.8 (0.3)	12.4 (0.2)	7.2 (0.3)	12.3 (0.1)
24	—	—	7.7* (0.3)	12.6 (0.1)	8.7 (0.6)	12.6 (0.2)	8.3 (0.4)	12.4 (0.2)	7.2 (0.4)	12.4 (0.3)
28	—	—	—	—	8.7 (0.4)	12.6 (0.2)	8.6 (0.4)	12.6 (0.2)	7.3 (0.4)	12.5 (0.2)



*Biological and ecological implications*

Commercially important North American shrimp of the genus *Penaeus* spawn at sea. As shown for *P. setiferus*, the larvae develop in the open sea, migrate into the estuarine areas as postlarvae, remain in the less saline estuaries until they approach maturity, and then return to the sea (Weymouth, Lindner and Anderson, 1933; Burkenroad, 1934; Pearson, 1939). Various studies in the field have suggested that postlarval and juvenile *Penaeus* are associated with low salinities characteristic of the estuary, and that postlarvae require the lowest salinity for growth and survival (Gunter, 1945, 1950; Pearse and Gunter, 1957). Lindner and Anderson (1956) concluded, however, that size of white shrimp (juveniles and subadults) seemed more closely related to locale than to salinity. Gunter, Christmas and Killebrew (1964) have recently presented additional field data indicating differences in the natural distributions, with respect to salinity, of the three commercial species, *P. aztecus*, *P. duorarum*, and *P. setiferus*. In so doing, these authors have made certain assumptions. For example (p. 184): "If salinity meant nothing to these animals they would be evenly distributed relatively over the whole range, if food were available. The general food habits of shrimp are still largely unknown, but all indications are they are omnivorous feeders, and shrimp do find food over the full salinity range up to pure sea water, although the food doubtless changes with size." The fact that shrimp do apparently eat a variety of food does not, however, indicate that all such foods are of comparable nutritive value (Williams, 1959; Zein-Eldin, 1963). Furthermore, there is no evidence that food is equally available throughout the salinity range occupied by shrimp in nature. Only in a previous study (Zein-Eldin, 1963) and in the work reported here, has food been equally available to all animals regardless of salinity. The 24-hour survival experiments, as well as the growth study, indicated that for *P. aztecus* postlarvae, only extreme salinity conditions influence growth and survival. Even normal oceanic salinity is not sufficient to interfere with postlarval brown shrimp growth and survival when other factors (temperature, food supply, predation, oxygen, light, pollution, etc.) are kept relatively constant. In view of our results, we suggest that other factors, such as food or cover (which may themselves require relatively narrow salinity ranges), are of greater importance than salinity *per se* in determining distribution, growth, and survival of these animals.

In the present studies, both the survival and the growth data indicated that wide ranges of salinity and temperature were well tolerated by postlarval brown shrimp. The combination of low salinity and low temperature, however, was not favorable, either for survival or growth. That *P. aztecus* can withstand extreme conditions of both factors has been demonstrated in the field as well, although published records have been largely limited to occurrences of juvenile and adult forms (Gunter, 1950). Bearden (1961), who found postlarval brown shrimp at temperatures as low as 6.5° C., noted a marked decrease in their abundance following the sudden cold spell which resulted in this low-temperature value. Renfro and Baxter (unpublished) have reported live postlarvae at 12° C. and 31.0‰ as well as at 2° C. and 30.5‰, supporting our laboratory evidence that low temperatures can be survived when salinities are sufficiently high. Comparable data of postlarval occurrence in low-salinity areas are not yet available. Brown shrimp (size not stated) have also been reported in salinities as low as 0.8‰



(Gunter and Shell, 1958) in Louisiana, while Gunter and Hall (1963) report a 34- and a 38-mm. specimen at 0.22‰ in the St. Lucie estuary in Florida. No temperatures were reported with the latter data, however. It must be noted, nevertheless, that St. Amant, Corkum and Broom (1963) reported maximal spring abundance of postlarval brown shrimp in Louisiana bays only after water temperatures consistently exceeded 20° C.

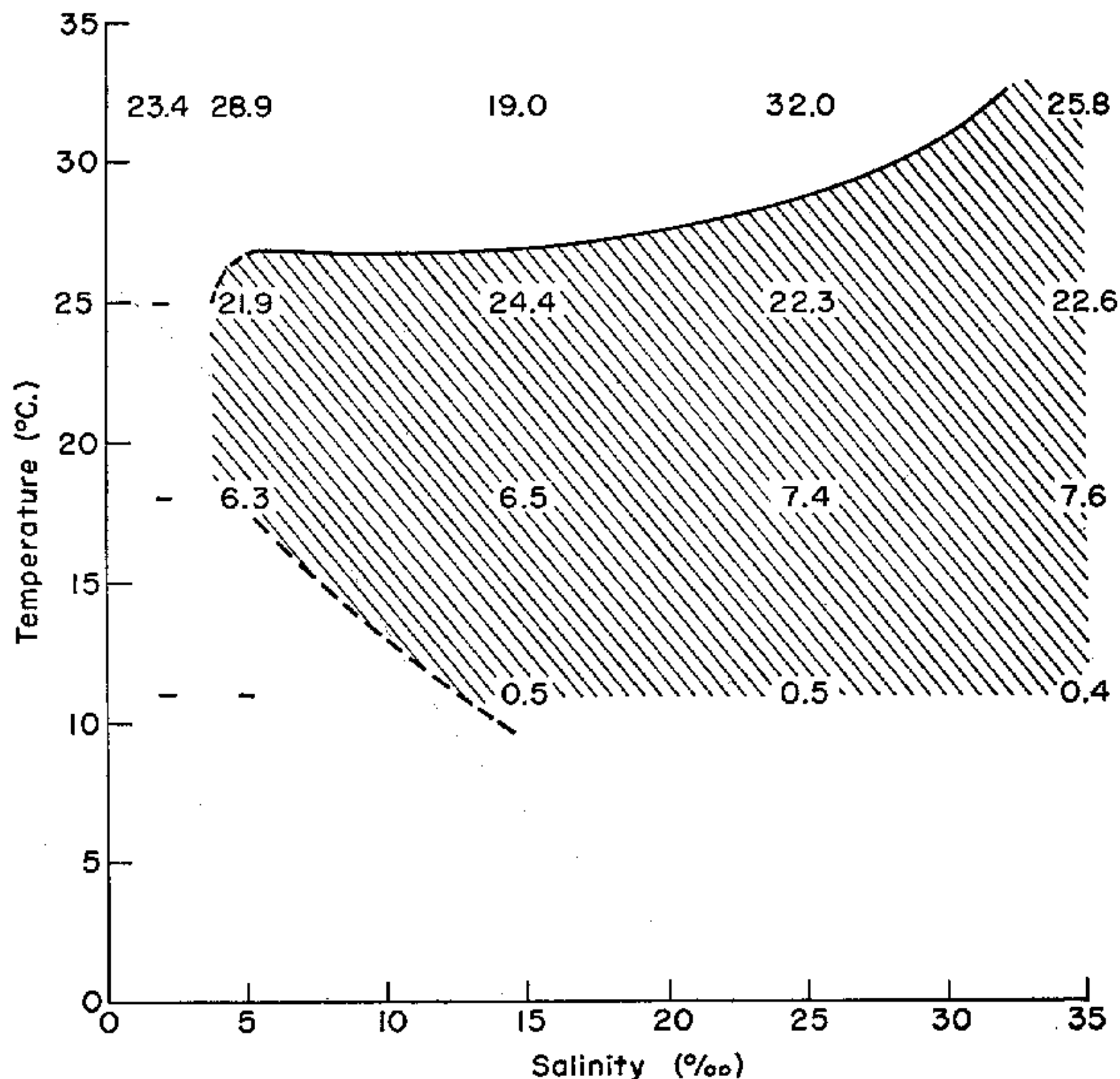


FIGURE 5. Growth and survival of young *P. aztecus* held 28 days at indicated levels of temperature and salinity. Numbers indicate increase in mean length (mm.). Hatched zone indicates 80-100% survival.

The temperature range permitting growth is more limited than the range for survival (Fig. 5). Our laboratory studies have demonstrated that growth can occur over a wide range of salinity at temperatures of 25° C. and above, and suggest that the effect of temperature upon the rate of growth increases rapidly with temperature between 11° and 25° C. (Fig. 6). This effect of temperature has been confirmed in more recent experiments in which we observed growth at a greater number of temperature levels between 15° and 35° C. than tested here.

The greatest growth differential per 7° C. was observed between the 18° and 25° C. levels. This difference may well explain the observation of St. Amant, Corkum and Broom (1963, p. 25) that "metamorphosis of postlarvae into rapidly growing juveniles occurs suddenly after water temperature exceeds 20° C." Above 25° C., increasing temperature has less effect upon growth. The recent experiments

referred to above indicate that growth is maximal at 30° to 32.5° C. This result, coupled with the increased mortality at 32° C., suggests that such a temperature condition, although promoting rapid growth in some individuals, may be above the optimum temperature for long-term growth and survival of *P. aztecus* postlarvae.

The laboratory evidence suggests that normal winter temperatures render the brackish bay systems unfavorable for both survival and growth of brown shrimp postlarvae, whereas almost any salinity will provide a favorable environment at normal summer temperatures. Thus, the pattern of tolerance to salinity and temperature observed in the laboratory may explain the seasonal distribution of *P.*

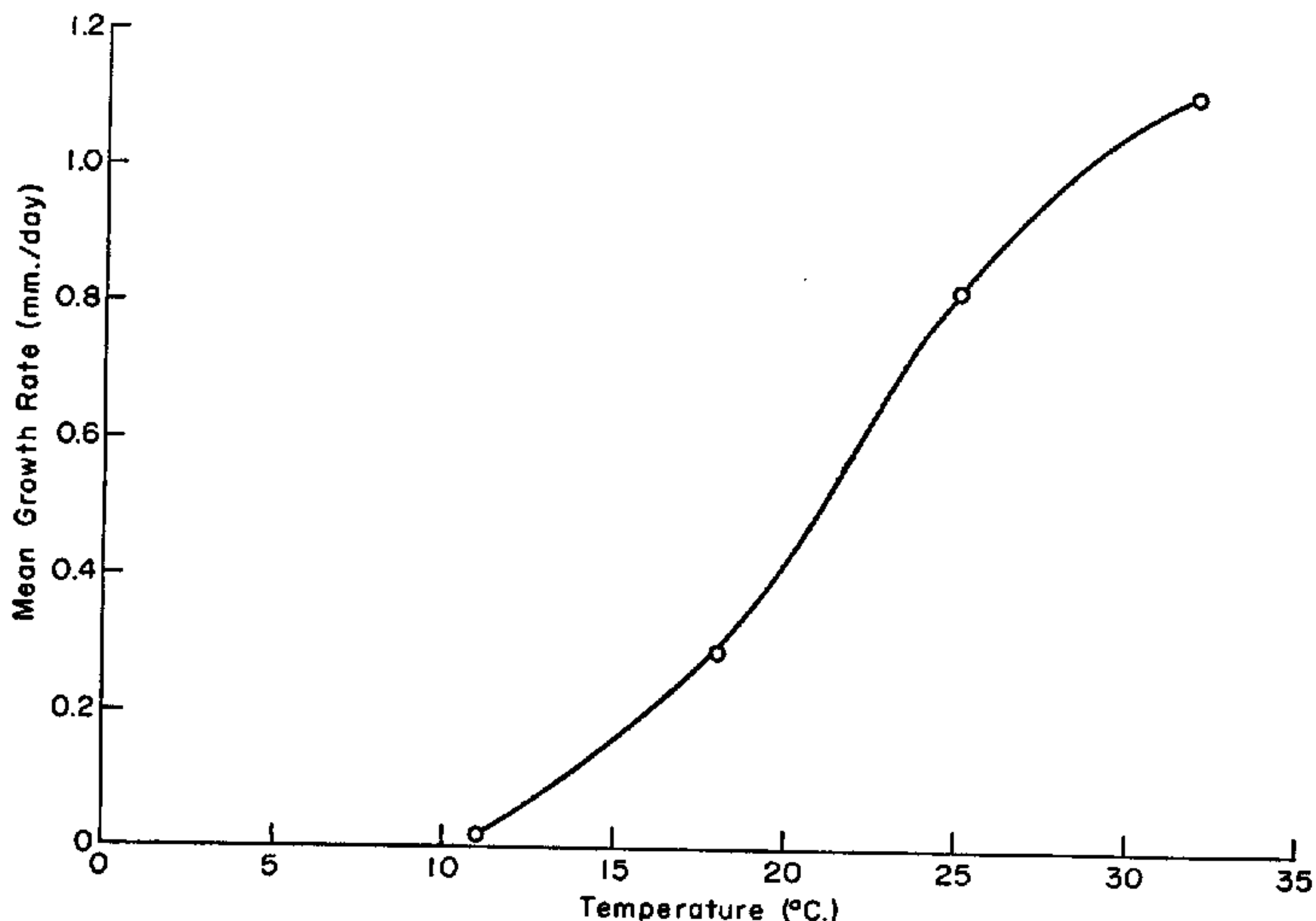


FIGURE 6. Effect of temperature on the laboratory growth rate of young *P. aztecus* (salinity: 25‰; length of experiment: 28 days; initial length of experimental shrimp: 12.1 mm.).

*aztecus* in much the same manner as described by Broekema (1941) for the migratory European shrimp, *Crangon crangon*. Survival of postlarvae within the estuary may also be affected by decreases in temperature or salinity. In the spring, postlarvae entering bays having relatively low temperatures and salinities above 15‰ may be adversely affected by a sudden salinity drop, such as that caused by heavy spring rains. Conversely, if the temperatures are intermediate (18° C., for example) but salinities low (10‰ or less), a drop in temperature may also decrease survival. Simultaneous decreases in both physical factors—temperature and salinity—would be most detrimental to the population in terms of both survival and growth.

Williams (1960) had previously noted the effects of temperature and salinity on juveniles and subadults of *P. aztecus*. Not only did he find that the 96-hour survival of 42- to 100-mm. specimens declined with decreasing temperature over

the range 28.8° C. to 8.8° C., but he also determined that survival was most markedly reduced at 10‰ (the lowest salinity tested) regardless of the temperature. Animals exposed to 8.8° C. showed a greater tendency to lose the ability to regulate the osmotic concentration of the serum. It is of interest that juveniles were better able to regulate serum concentration than were adults (120 to 150 mm.) exposed to the same conditions. McFarland and Lee (1963) demonstrated that brown shrimp adults were better osmoregulators at higher salinity than at lower, with a greater tendency to isosmoticity when the external medium was below 18‰. The latter authors were unable to study animals in salinities below 5‰ to 6‰ since only one of 12 adults survived 24-hour exposure to this range of salinity, despite an acclimation period of almost one week.

The studies cited above suggest that salinity tolerance may vary not only with temperature, but also with size (age) of the shrimp. In demonstrating good survival of *P. aztecus* postlarvae over a broad range of salinity and temperature, the findings presented here suggest that postlarvae of this species are better osmoregulators than juveniles, which were tested by Williams (1960), or the adults tested by McFarland and Lee (1963). Further studies are planned to determine the effects of both temperature and salinity upon the osmotic behavior in various life-history stages of *P. aztecus* and *P. setiferus*.

#### SUMMARY

1. The combined effects of salinity and temperature upon growth and survival of postlarvae of the brown shrimp, *Penaeus aztecus*, were studied under controlled conditions.

2. Test salinity ranged from 2‰ to 40‰ and temperature from 7° to 35° C.

3. With relatively short periods of acclimation, postlarval brown shrimp withstood wide fluctuations in both temperature and salinity for 24 hours.

4. The range of tolerance to these factors over periods of 28 days was only slightly less than that observed for 24 hours.

5. Postlarvae survived temperatures as low as 11° C. with almost no growth for one month in salinities of about 15‰ or above.

6. Growth increased with temperature, with significant growth beginning at some temperature above 11° C. but below 18° C. The most marked increase in growth rate occurred in the temperature region between 11° and 25° C.

7. At temperatures below 15° C., young (postlarval) shrimp demonstrated a decreased tolerance to low salinity. This reduced tolerance may influence the natural distribution and survival of postlarvae, which do not ordinarily enter the estuaries in abundance until spring when the temperature has increased to levels at which characteristically low estuarine salinities are no longer harmful.

8. Salinity *per se* had little effect on either survival or growth, except at extreme temperatures.

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